

WEST Search History

DATE: Thursday, February 13, 2003

<u>Set Name</u>	<u>Query</u>	<u>Hit Count</u>	<u>Set Name</u>
side by side			result set

DB=USPT,DWPI; PLUR=YES; OP=ADJ

L1	erikson-G\$.in. or Daksis-J\$.in.	12	L1
L2	5030557	98	L2
L3	enhanc\$ near10 hybridiz\$	1179	L3
L4	hairpin near5 block\$ agent	0	L4
L5	hairpin near block\$	9	L5
L6	hairpin same block\$	372	L6
L7	probe same target same hybridiz\$	6923	L7
L8	L7 and l3	687	L8
L9	L8 and l1	1	L9
L10	L8 and l6	16	L10
L11	6265170.pn. or 5770365.pn.	4	L11

END OF SEARCH HISTORY

Connecting via Winsock to STN

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LOGINID:SSSPTA1655CXW

PASSWORD:

TERMINAL (ENTER 1, 2, 3, OR ?):2

* * * * * Welcome to STN International * * * * *

NEWS	1		Web Page URLs for STN Seminar Schedule - N. America
NEWS	2	Apr 08	"Ask CAS" for self-help around the clock
NEWS	3	Apr 09	BEILSTEIN: Reload and Implementation of a New Subject Area
NEWS	4	Apr 09	ZDB will be removed from STN
NEWS	5	Apr 19	US Patent Applications available in IFICDB, IFIPAT, and
IFIUDB			
NEWS	6	Apr 22	Records from IP.com available in CAPLUS, HCAPLUS, and
ZCAPLUS			
NEWS	7	Apr 22	BIOSIS Gene Names now available in TOXCENTER
NEWS	8	Apr 22	Federal Research in Progress (FEDRIP) now available
NEWS	9	Jun 03	New e-mail delivery for search results now available
NEWS	10	Jun 10	MEDLINE Reload
NEWS	11	Jun 10	PCTFULL has been reloaded
NEWS	12	Jul 02	FOREGE no longer contains STANDARDS file segment
NEWS	13	Jul 22	USAN to be reloaded July 28, 2002; saved answer sets no longer valid
NEWS	14	Jul 29	Enhanced polymer searching in REGISTRY
NEWS	15	Jul 30	NETFIRST to be removed from STN
NEWS	16	Aug 08	CANCERLIT reload
NEWS	17	Aug 08	PHARMAMarketLetter(PHARMAML) - new on STN
NEWS	18	Aug 08	NTIS has been reloaded and enhanced
NEWS	19	Aug 19	Aquatic Toxicity Information Retrieval (AQUIRE) now available on STN
NEWS	20	Aug 19	IFIPAT, IFICDB, and IFIUDB have been reloaded
NEWS	21	Aug 19	The MEDLINE file segment of TOXCENTER has been reloaded
NEWS	22	Aug 26	Sequence searching in REGISTRY enhanced
NEWS	23	Sep 03	JAPIO has been reloaded and enhanced
NEWS	24	Sep 16	Experimental properties added to the REGISTRY file
NEWS	25	Sep 16	CA Section Thesaurus available in CAPLUS and CA
NEWS	26	Oct 01	CASREACT Enriched with Reactions from 1907 to 1985
NEWS	27	Oct 21	EVENTLINE has been reloaded
NEWS	28	Oct 24	BEILSTEIN adds new search fields
NEWS	29	Oct 24	Nutraceuticals International (NUTRACEUT) now available on
STN			
NEWS	30	Oct 25	MEDLINE SDI run of October 8, 2002
NEWS	31	Nov 18	DKILIT has been renamed APOLLIT
NEWS	32	Nov 25	More calculated properties added to REGISTRY
NEWS	33	Dec 02	TIBKAT will be removed from STN
NEWS	34	Dec 04	CSA files on STN
NEWS	35	Dec 17	PCTFULL now covers WP/PCT Applications from 1978 to date
NEWS	36	Dec 17	TOXCENTER enhanced with additional content
NEWS	37	Dec 17	Adis Clinical Trials Insight now available on STN
NEWS	38	Dec 30	ISMEC no longer available
NEWS	39	Jan 13	Indexing added to some pre-1967 records in CA/CAPLUS
NEWS	40	Jan 21	NUTRACEUT offering one free connect hour in February 2003

NEWS 41 Jan 21 PHARMAML offering one free connect hour in February 2003
NEWS 42 Jan 29 Simultaneous left and right truncation added to COMPENDEX,
ENERGY, INSPEC

NEWS EXPRESS January 6 CURRENT WINDOWS VERSION IS V6.01a,
CURRENT MACINTOSH VERSION IS V6.0b(ENG) AND V6.0Jb(JP),
AND CURRENT DISCOVER FILE IS DATED 01 OCTOBER 2002
NEWS HOURS STN Operating Hours Plus Help Desk Availability
NEWS INTER General Internet Information
NEWS LOGIN Welcome Banner and News Items
NEWS PHONE Direct Dial and Telecommunication Network Access to STN
NEWS WWW CAS World Wide Web Site (general information)

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* * * * * STN Columbus * * * * *

FILE 'HOME' ENTERED AT 13:09:57 ON 13 FEB 2003

=> file .biotech caplus
COST IN U.S. DOLLARS

SINCE FILE	TOTAL
ENTRY	SESSION
1.47	1.47

FULL ESTIMATED COST

FILES 'MEDLINE, BIOTECHDS, EMBASE, BIOSIS, SCISEARCH, CANCERLIT, CAPLUS'
ENTERED AT 13:13:57 ON 13 FEB 2003
ALL COPYRIGHTS AND RESTRICTIONS APPLY. SEE HELP USAGETERMS FOR DETAILS.

7 FILES IN THE FILE LIST

=> search Erikson G?/au or Daksis J?/au
L1 134 ERIKSON G?/AU OR DAKSIS J?/AU

=> s enhanc? and hybridiz
=> s enhanc (P) hybridiz?
PROXIMITY OPERATOR LEVEL NOT CONSISTENT WITH
FIELD CODE - 'AND' OPERATOR ASSUMED 'ENHANC (P) HYBRIDIZ?'
L2 0 ENHANC (P) HYBRIDIZ?

=> s enhanc? and hybridiz?
L3 29614 ENHANC? AND HYBRIDIZ?

=> s hairpin (s) block###
L4 752 HAIRPIN (S) BLOCK###

=> s helper (s) oligonucl?
L5 697 HELPER (S) OLIGONUCL?

=> s probe (s) target(s) hybridiz?
L6 6618 PROBE (S) TARGET(S) HYBRIDIZ?

=> d his

(FILE 'HOME' ENTERED AT 13:09:57 ON 13 FEB 2003)

FILE 'MEDLINE, BIOTECHDS, EMBASE, BIOSIS, SCISEARCH, CANCERLIT, CAPLUS'
ENTERED AT 13:13:57 ON 13 FEB 2003

L1 134 SEARCH ERIKSON G?/AU OR DAKSIS J?/AU
L2 0 S ENHANC (P) HYBRIDIZ?
L3 29614 S ENHANC? AND HYBRIDIZ?
L4 752 S HAIRPIN (S) BLOCK###
L5 697 S HELPER (S) OLIGONUCL?
L6 6618 S PROBE (S) TARGET(S) HYBRIDIZ?

=> s l1 and l3

L7 4 L1 AND L3

=> s l3 and l6

L8 462 L3 AND L6

=> s l8 and (l4 or l5)

L9 10 L8 AND (L4 OR L5)

=> d his

(FILE 'HOME' ENTERED AT 13:09:57 ON 13 FEB 2003)

FILE 'MEDLINE, BIOTECHDS, EMBASE, BIOSIS, SCISEARCH, CANCERLIT, CAPLUS'
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L1 134 SEARCH ERIKSON G?/AU OR DAKSIS J?/AU
L2 0 S ENHANC (P) HYBRIDIZ?
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L6 6618 S PROBE (S) TARGET(S) HYBRIDIZ?
L7 4 S L1 AND L3
L8 462 S L3 AND L6
L9 10 S L8 AND (L4 OR L5)

=> d ibib abs l7 1-4

L7 ANSWER 1 OF 4 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.

ACCESSION NUMBER: 2001:440564 BIOSIS

DOCUMENT NUMBER: PREV200100440564

TITLE: Homogenous assay of duplex of triplex **hybridization**
by means of multiple measurements under varied

conditions.

AUTHOR(S): Picard, Pierre (1); **Daksis, Jasmine I.;**
Erikson, Glen H.

CORPORATE SOURCE: (1) Brampton Canada

ASSIGNEE: Ingeneus Corporation, Bridgetown, Barbados

PATENT INFORMATION: US 6265170 July 24, 2001

SOURCE: Official Gazette of the United States Patent and Trademark
Office Patents, (July 24, 2001) Vol. 1248, No. 4, pp. No
Pagination. e-file.
ISSN: 0098-1133.

DOCUMENT TYPE: Patent

LANGUAGE: English

AB The invention provides homogeneous assay methods for nucleic acid
hybridization, detection and evaluation. The assay includes
obtaining signals from a test sample both before and during the
application of a voltage to the test sample and correlating the signals,

each of which is indicative of the binding affinity of the probe and the target to each other. The assay enables determining an extent of matching between the probe and the target, as the voltage can be calibrated so as to destabilize significantly any **hybridization** except perfectly complementary **hybridization**. The signals whose magnitude is correlated with binding affinity can be electrical conductance and/or fluorescent intensity. Preferably, both signal pairs are measured and compared so as to **enhance** the reliability of the assay. The assay can detect specific **hybridization** between single-stranded probes and non-denatured double-stranded targets to form triplexes, thus obviating the need to denature the targets. The assay methods can also be applied to duplex **hybridization** complexes.

L7 ANSWER 2 OF 4 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 2002:736774 CAPLUS

DOCUMENT NUMBER: 137:258475

TITLE: Homogeneous assay of nucleic acid
hybridization by means of multiple
measurements under varied conditions

INVENTOR(S): **Erikson, Glen H.; Daksis, Jasmine I.**
; Picard, Pierre

PATENT ASSIGNEE(S): Turks/Caicos I.

SOURCE: U.S. Pat. Appl. Publ., 22 pp., Cont.-in-part of U.S.
6,265,170.

CODEN: USXXCO

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 3

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 2002137056	A1	20020926	US 2001-911047	20010723
US 6265170	B1	20010724	US 2000-490273	20000124
US 2002123066	A1	20020905	US 2002-120092	20020410
WO 2003010506	A2	20030206	WO 2002-IB2788	20020715

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN,
CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH,
GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR,
LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH,
PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ,
UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU,
TJ, TM

RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, BG,
CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL,
PT, SE, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR,
NE, SN, TD, TG

PRIORITY APPLN. INFO.: US 2000-490273 A2 20000124
US 2001-911047 A2 20010723

AB A method for homogeneously assaying biopolymer bonding, specifically nucleic acid **hybridization**, includes obtaining signals from a test sample before, during and/or after the application of stimulus to the test sample and correlating the signals. The signals, whose magnitude correlate with binding affinity, can be, for example, elec. conductance and/or fluorescent intensity. The stimulus can be, for example, elec. voltage and/or laser radiation. Preferably, different types of signals are measured and compared so as to **enhance** the reliability of the assay.

L7 ANSWER 3 OF 4 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 2002:696538 CAPLUS
 DOCUMENT NUMBER: 137:227601
 TITLE: Nucleic acid binding **enhancement** by
 conjugation with nucleotides, nucleosides, bases
 and/or their analogs for improved degree and
 specificity of **hybridization**
 INVENTOR(S): Erikson, Glen H.; Daksis, Jasmine I.
 PATENT ASSIGNEE(S): Ingeneus Corporation, Barbados
 SOURCE: U.S. Pat. Appl. Publ., 9 pp., Cont.-in-part of U. S.
 Ser. No. 909,496.
 CODEN: USXXCO
 DOCUMENT TYPE: Patent
 LANGUAGE: English
 FAMILY ACC. NUM. COUNT: 7
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE																		
US 2002127590	A1	20020912	US 2002-80767	20020222																		
US 6403313	B1	20020611	US 1999-468679	19991221																		
US 6420115	B1	20020716	US 2000-613263	20000710																		
US 2002031775	A1	20020314	US 2001-909496	20010720																		
WO 2002024946	A2	20020328	WO 2001-IB1643	20010910																		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG																						
AU 2001084350	A5	20020402	AU 2001-84350	20010910																		
PRIORITY APPLN. INFO.: <table border="0" style="margin-left: 400px;"> <tr> <td>US 1999-468679</td> <td>A2</td> <td>19991221</td> </tr> <tr> <td>US 2000-613263</td> <td>A2</td> <td>20000710</td> </tr> <tr> <td>US 2000-664827</td> <td>A2</td> <td>20000919</td> </tr> <tr> <td>US 2001-281547P</td> <td>P</td> <td>20010404</td> </tr> <tr> <td>US 2001-909496</td> <td>A2</td> <td>20010720</td> </tr> <tr> <td>WO 2001-IB1643</td> <td>W</td> <td>20010910</td> </tr> </table>					US 1999-468679	A2	19991221	US 2000-613263	A2	20000710	US 2000-664827	A2	20000919	US 2001-281547P	P	20010404	US 2001-909496	A2	20010720	WO 2001-IB1643	W	20010910
US 1999-468679	A2	19991221																				
US 2000-613263	A2	20000710																				
US 2000-664827	A2	20000919																				
US 2001-281547P	P	20010404																				
US 2001-909496	A2	20010720																				
WO 2001-IB1643	W	20010910																				
AB An improved method of forming a specific complex between a probe contg. probe nucleobases and a target contg. target nucleobases, includes mixing the probe and the target under hybridizing conditions, wherein the probe and/or the target is conjugated to a blocking agent, which enhances the avidity and/or specificity of hybridization , whether by Watson-Crick motif or by homologous binding motif. The blocking agent contains at least one nucleobase and can be, e.g., a free nucleobase, a nucleoside or a nucleotide. Conjugation enhances hybridization by hindering the probe and/or target from existing in a conformation antithetical to hybridization . Thus, reaction of 2 pmoles of a 15-mer single-stranded DNA probe contg. 6 adenine bases (conjugated with 3 pmoles of thymine) with 2 pmoles of a wild-type antisense strand (50-mer) in the presence of YOYO-1 results in 78% increased formation of parallel homologous complexes between perfectly homologous sequences. By contrast, the efficiency of formation of parallel homologous complexes contg. a 1-bp A-G mismatch was increased by only 16% when the probe was conjugated 25% with thymine.																						
L7 ANSWER 4 OF 4 CAPLUS COPYRIGHT 2003 ACS ACCESSION NUMBER: 2001:537425 CAPLUS DOCUMENT NUMBER: 135:133083 TITLE: Homogenous assay of duplex of triplex																						

INVENTOR(S) : **hybridization** by means of multiple
measurements under varied conditions
Picard, Pierre; **Daksis, Jasmine I.;**
Erikson, Glen H.
PATENT ASSIGNEE(S) : Ingeneus Corporation, Barbados
SOURCE : U.S., 22 pp.
CODEN: USXXAM
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 3
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 6265170	B1	20010724	US 2000-490273	20000124
WO 2001053526	A2	20010726	WO 2001-IB77	20010123
WO 2001053526	A3	20020613		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
EP 1250459	A2	20021023	EP 2001-902567	20010123
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
US 2002137056	A1	20020926	US 2001-911047	20010723
US 2002123066	A1	20020905	US 2002-120092	20020410
PRIORITY APPLN. INFO.:			US 2000-490273	A 20000124
			WO 2001-IB77	W 20010123
			US 2001-911047	A2 20010723

AB The invention provides homogeneous assay methods for nucleic acid **hybridization**, detection and evaluation. The assay includes obtaining signals from a test sample both before and during the application of a voltage to the test sample and correlating the signals, each of which is indicative of the binding affinity of the probe and the target to each other. The assay enables detg. an extent of matching between the probe and the target, as the voltage can be calibrated so as to destabilize significantly any **hybridization** except perfectly complementary **hybridization**. The signals whose magnitude is correlated with binding affinity can be elec. conductance and/or fluorescent intensity. Preferably, both signal pairs are measured and compared so as to **enhance** the reliability of the assay. The assay can detect specific **hybridization** between single-stranded probes and non-denatured double-stranded targets to form triplexes, thus obviating the need to denature the targets. The assay methods can also

be applied to duplex **hybridization** complexes. The invention can discriminate between perfectly matched dsDNA:PNA triplex hybrids and those contg. 1 bp or 2 bp mismatches. Intercalation by YOYO-1 facilitated the formation of the dsDNA-PNA triplexes and dsDNA-ssDNA triplexes.

REFERENCE COUNT: 18 THERE ARE 18 CITED REFERENCES AVAILABLE FOR THIS

FORMAT RECORD. ALL CITATIONS AVAILABLE IN THE RE

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(FILE 'HOME' ENTERED AT 13:09:57 ON 13 FEB 2003)

FILE 'MEDLINE, BIOTECHDS, EMBASE, BIOSIS, SCISEARCH, CANCERLIT, CAPLUS'
ENTERED AT 13:13:57 ON 13 FEB 2003

L1 134 SEARCH ERIKSON G?/AU OR DAKSIS J?/AU
L2 0 S ENHANC (P) HYBRIDIZ?
L3 29614 S ENHANC? AND HYBRIDIZ?
L4 752 S HAIRPIN (S) BLOCK###
L5 697 S HELPER (S) OLIGONUCL?
L6 6618 S PROBE (S) TARGET(S) HYBRIDIZ?
L7 4 S L1 AND L3
L8 462 S L3 AND L6
L9 10 S L8 AND (L4 OR L5)

=> d ibib abs l9 1-10

L9 ANSWER 1 OF 10 MEDLINE
ACCESSION NUMBER: 2001075808 MEDLINE
DOCUMENT NUMBER: 20378673 PubMed ID: 10919826
TITLE: Unlabeled **helper oligonucleotides**
increase the in situ accessibility to 16S rRNA of
fluorescently labeled **oligonucleotide** probes.
AUTHOR: Fuchs B M; Glockner F O; Wulf J; Amann R
CORPORATE SOURCE: Max-Planck-Institut fur Marine Mikrobiologie, D-28359
Bremen, Germany.. bfuchs@mpi-bremen.de
SOURCE: APPLIED AND ENVIRONMENTAL MICROBIOLOGY, (2000 Aug) 66 (8)
3603-7.
Journal code: 7605801. ISSN: 0099-2240.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200101
ENTRY DATE: Entered STN: 20010322
Last Updated on STN: 20010322
Entered Medline: 20010104

AB **Target** site inaccessibility represents a significant problem for
fluorescence in situ **hybridization** (FISH) of 16S rRNA with
oligonucleotide probes. Here, unlabeled **oligonucleotides**
(helpers) that bind adjacent to the **probe target** site
were evaluated for their potential to increase weak **probe**
hybridization signals in Escherichia coli DSM 30083(T). The use of
helpers **enhanced** the fluorescence signal of all six probes
examined at least fourfold. In one case, the signal of **probe**
Eco474 was increased 25-fold with the use of a single **helper**
probe, H440-2. In another case, four unlabeled helpers raised the
FISH signal of a formerly weak **probe**, Eco585, to the level of
the brightest monolabeled **oligonucleotide** probes available for
E. coli. The temperature of dissociation and the mismatch discrimination
of probes were not significantly influenced by the addition of helpers.
Therefore, using helpers should not cause labeling of additional
nontarget
organisms at a defined stringency of **hybridization**. However, the
helper action is based on sequence-specific binding, and there is
thus a potential for narrowing the **target** group which must be
considered when designing helpers. We conclude that helpers can open
inaccessible rRNA regions for FISH with **oligonucleotide** probes
and will thereby further improve the applicability of this technique for
in situ identification of microorganisms.

a probe to the 430-500 region of Salmonella 16 S rRNA, as well as helper oligonucleotides I and/or II. In an overnight hybridization protocol using probe alone, probe/I, probe/II, or probe/I/II, percent hybrid found was 1.8, 68.5, 68.0, or 83.1%, resp.

The

Tm of the probe alone, probe/I, and probe/II were 59.degree., 63.5.degree., and 63.0.degree., resp. A kit contg. helper oligonucleotides is claimed.

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L3	29614 S ENHANC? AND HYBRIDIZ?
L4	752 S HAIRPIN (S) BLOCK###
L5	697 S HELPER (S) OLIGONUCL?
L6	6618 S PROBE (S) TARGET(S) HYBRIDIZ?
L7	4 S L1 AND L3
L8	462 S L3 AND L6
L9	10 S L8 AND (L4 OR L5)

=> end

ALL L# QUERIES AND ANSWER SETS ARE DELETED AT LOGOFF

LOGOFF? (Y)/N/HOLD:y

COST IN U.S. DOLLARS

SINCE FILE	TOTAL
ENTRY	SESSION
74.64	76.11

FULL ESTIMATED COST

DISCOUNT AMOUNTS (FOR QUALIFYING ACCOUNTS)

SINCE FILE	TOTAL
ENTRY	SESSION
-4.56	-4.56

CA SUBSCRIBER PRICE

STN INTERNATIONAL LOGOFF AT 13:28:35 ON 13 FEB 2003

L9 ANSWER 2 OF 10 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI

ACCESSION NUMBER: 2002-16605 BIOTECHDS

TITLE: Novel **oligonucleotides** functioning as **hybridization** probes, **helper** probes and/or primers, targeted to nucleic acid sequences derived from *Cryptosporidium* organisms, useful for detecting the organism in a test sample; involving DNA probe, DNA primer and **hybridization** for use in protozoan detection in water, excrement and food samples

AUTHOR: CUNNINGHAM M M; STULL P D; WEISBURG W G

PATENT ASSIGNEE: GEN-PROBE INC

PATENT INFO: WO 2002022890 21 Mar 2002

APPLICATION INFO: WO 2000-US42192 12 Sep 2000

PRIORITY INFO: US 2000-232028 12 Sep 2000

DOCUMENT TYPE: Patent

LANGUAGE: English

OTHER SOURCE: WPI: 2002-454395 [48]

AN 2002-16605 BIOTECHDS

AB DERWENT ABSTRACT:

NOVELTY - **Oligonucleotides** functioning as **hybridization** assay probes, **helper** probes and/or amplification primers, targeted to nucleic acid sequences derived from *Cryptosporidium* (Cp) organisms, are new.

DETAILED DESCRIPTION - The **oligonucleotide** may be: (a) **hybridization** assay **probe** (I) comprising an **oligonucleotide** which **hybridizes** to a **target** sequence present in nucleic acid derived from Cp organism or *C.parvum* in a test sample under stringent conditions to form (I):**target** hybrid stable for detection, where (I) has an at least 10 contiguous

base

region that is 80% complementary to an at least 10 contiguous base

region

present in the **target** sequence e.g. (S1-S4) (all of which are derived from a Cp organism) or (S5-S20) (all of which are derived from *C.parvum*), where the **probe** complementary to sequence of (S1)-(S4) does not **hybridize** to nucleic acid derived from non-Cp organism, and the **probe** complementary to sequence of (S5)-(S20) does not **hybridize** to a nucleic acid derived from *C.muris*, *C.baileyi*, *C.wrairi*, in the test sample, to form **probe** :non-**target** hybrid stable for detection under stringent conditions; (b) a **probe** mix (II) comprising (I) and one or more **helper oligonucleotides** with an at least 10 contiguous base region which is at least 80% complementary to an at least 10 contiguous base region present in a **target** sequence e.g. (S21-S44), where (S21-29) are derived from a Cp organism and (S30-44)

are

derived from a *C.parvum* organism; or (c) an amplification primer (III) for use in amplifying a nucleic acid sequence present in nucleic acid derived from Cp organism under amplification conditions, where the

primer

comprises an **oligonucleotide** with at least 10 contiguous base region that is at least 80% complementary to at least 10 contiguous base region present in a **target** sequence e.g. (S45-S68) (the amplification primer is directed to 18S ribosomal nucleic acid derived from Cp or *C.parvum* organism). INDEPENDENT CLAIMS are also included for: (1) a set of amplification primers (IV) for use in amplifying a nucleic acid sequences present in nucleic acid derived from Cp organisms under amplification conditions, where at least two primers of the set of primers comprises an **oligonucleotide** with at least 10

contiguous base region that is at least 80% complementary to an at least 10 contiguous base region present in **target** sequence of (S45)-(S68), where the primers of the set of primers optionally include

a

5' sequence that is recognized by an RNA polymerase or **enhances** initiation or elongation by a RNA polymerase; (2) an **oligonucleotide** (V) for use in determining the presence of a *C. parvum* organism in a test sample, the **oligonucleotide** with at least 10 contiguous base region that is at least 80% complementary to an at least 10 contiguous base region present in **target** sequence of (S1)-(S20), (S45)-(S67) or (S68), where the **oligonucleotide** optionally includes a 5' sequence which is recognized by an RNA polymerase or which **enhances** initiation or elongation by RNA polymerase; (3) a kit (VI) comprising, in packaged combination two or more (V); (4) a kit (VII) comprising in packaged combination (I) and at least one **helper oligonucleotide** with at least 10 contiguous base region that is at least 80% complementary to an at least 10 contiguous base region present in **target** sequence of (S21)-(S44); (5) a kit (VIII) comprising in packaged combination, (I)

and

(III); (6) a kit (IX) comprising in packaged combination, two or more amplification primers, where at least two of the primers are selected from (IV); and (7) obtaining (M1) purified RNA from a viable oocyst comprises (a) centrifuging a fluid sample suspected of containing

oocysts

at a speed and for a period of time sufficient to concentrate the

oocysts

within a vessel containing the fluid sample; (b) removing a supernatant from the vessel; (c) resuspending the concentrated oocysts, if present, in a buffered solution; (d) agitating the buffered solution in the presence of several particles at a rate and for a period of time sufficient to lyse the oocysts and release RNA from them; (e) immobilizing the released RNA on a RNA-binding filter; (f) purifying the released RNA by washing the filter one or more times with a buffered solution to remove oocyst components other than the released RNA; and

(g)

removing the purified RNA from the filter. CTATCAGCTTTAGACGGTAGGG

(S1) CUAUCAGCUUUAGACGGUAGGG

(S2)

CCCTACCGTCTAAAGCTGATAG

(S3)

CCCUACCGUCUAAAGCUGAUAG

(S4)

GCGAAAAAAGCTGACTTTATGGAAGGG

(S5) GTTAAAGACAACTAATGCGAAAGC

(S8) GUUAAAGACAAACUAAUGCGAAAGC

(S12)

CAACCCTTCCATAAAGTCGAGTTTT

(S15)

GCUUUCGCAUUAAGUUUGUCUUUAC

(S20) GACATATCATTCAAGTTTCTGAC

(S21) GUCAGAAACUUGAUGAUUGUC

(S27)

GGAUAACCGUGGUAUUCUAGAGCUAAUACAU (S32) UAUUUGGUUCUUUAUCUAAUAAUACAA

(S44) GCCATGCATGTCTAAGTATAAAC

(S45)

GCCAAGGATGTTTTTCATTAATC

(S50)

GUAUUUACAGUCAGAGGUG

(S55)

CTGCCTTCCTTAGATGTGGTAG

(S60)

CCGUAAAGUUAUUAUGAGUCACC

(S65) GAUUAUGAAACAUCUUGGC

(S68)

WIDER DISCLOSURE - The following are disclosed: (1) compositions comprising a nucleic acid hybrid found between (I) and **target** nucleic acid under stringent **hybridization** assay conditions; (2) compositions comprising nucleic acid hybrids formed between the **helper probe** and **target** nucleic acid under stringent **hybridization** assay conditions; (3) a nucleic acid hybrid formed between an amplification primer and **target** nucleic acid under amplification conditions; (4) compositions for determining the presence or amount of a *Cp* organism in test sample

comprising nucleic acid hybrid derived from Cp organism and a **hybridization assay probe** comprising **oligonucleotide** with or substantially corresponding to base sequence of (S1)-(S4); (5) compositions for determining presence of an amount of C.parvum organism present in test sample comprising nucleic acid hybrid formed between **target** nucleic acid derived from C.parvum organism and **hybridization assay probe** which comprises an **oligonucleotide** with or substantially corresponding to sequence of (S5)-(S20); and (6) compositions for amplifying **target** sequences present in **target** nucleic acid derived from Cp, where the compositions comprise nucleic acid

hybrid

formed between a **target** nucleic acid and amplification primer comprising an **oligonucleotide** with or substantially corresponding to base sequence of (S45)-(S67) or (S68).

BIOTECHNOLOGY - Preferred **Oligonucleotide**: (I) or (III) comprises an **oligonucleotide** with at least 10 contiguous base region that is at least 90% (preferably 100%) complementary to an at least 10 contiguous base region present is the **target** sequence. Preferably, the base sequence of the **oligonucleotide** is 80% complementary to the base sequence of the **target** sequence, and thus the base sequence of the **probe** or primer is at least 80% complementary to the base sequence of the **target** sequence. Optionally, the base sequence of the **probe** or primer is fully complementary to the base sequence of the **target** sequence. (I) is up to 100 bases in length, preferably 12-50 (more preferably, 18-35) bases in length. The **probe** contains base sequences that **hybridize** to each other when not **hybridized** to the **target** sequence under the stringent conditions. Optionally, the **probe** comprises one or more base sequences which do not stably **hybridize** to nucleic acid derived from Cp organisms, or to nucleic acid derived from a non-**target** organism present in the test sample under stringent conditions, where the **probe** **hybridizes** to two of the one or more base sequences, where the two base sequences **hybridize** to each other when the **probe** is not **hybridized** to the **target** sequence under the stringent conditions. (I) further comprises a detectable label of a group of interacting labels including a

luminescent

or a quencher label. The **oligonucleotides** include at least one ribonucleotide modified to include 2'-O-methyl substitution to the ribofuranosyl moiety, and a pseudo peptide backbone joins at least a portion of the bases of the **oligonucleotide**. (III) is 18-40 bases in length and optionally includes the 5' sequence which is recognized by an RNA polymerase or which **enhances** initiation or elongation by an RNA polymerase. Preferably, the primer includes a 5' sequence described above. The 5' sequence which is recognized by RNA polymerase or which **enhances** initiation or elongation by RNA polymerase is a T7 promoter (P1). The primer optionally contains base sequences which **hybridize** to each other when not **hybridized** to the **target** sequence under the amplification conditions. The primer further includes a group of interacting labels as described above. (IV) includes first and second primers whose **target** sequences are any one of (S45)-(S68). The base sequence of (V) is at least 80% complementary (fully complementary) to the base sequence of the **target** sequence. AATTTAATACGACTCACTATAGGGAGA (P1) Preferred Kit: (VI) comprises two **oligonucleotides**, where the first **oligonucleotide** is complementary to a **target** sequence of any one of (S1)-(S20) and a second **oligonucleotide** complementary to a **target** sequence of (S45)-(S67) or (S68). (VIII) further comprises at least one

helper oligonucleotide with at least 10 contiguous base region which is at least 80% complementary to 10 contiguous base region in a **target** sequence of (S21)-(S43) or (S44). Preferred Method: In (M1), the agitating step includes oscillating the vessel containing the buffered solution. The particles have a spherical shape and an average diameter in the range of about 0.1-2.5 mm.

USE - (I) (complementary to a **target** sequence of (S1)-(S4)) is useful for determining the presence of Cp organism in a test sample, where the formation of (I):**target** hybrid is determined, and (I) complementary to **target** sequence of (S5)-(S20) is useful determining presence of C.parvum in a sample. For detecting C.parvum, the method further involves providing (III). (II) or (IV) is useful for amplifying Cp nucleic acid which may be present in a test sample. The method further involves determining the presence of amplified **target** sequence in test sample with a **hybridization assay probe** (claimed). (I) is useful for determining the presence of Cp organisms in general, and C.parvum organisms in particular in test samples of water, feces, food or other sample media.

ADVANTAGE - The **hybridization** assay probes are able to distinguish Cp and also C.parvum nucleic acids from non-Cp nucleic acids because of the ability of the **probe** to differentially **hybridize** to Cp nucleic acids under stringent **hybridization** assay conditions.

EXAMPLE - Amplification of a **target** sequence of Cryptosporidium rRNA and detection of the amplified rRNA using a **hybridization assay probe** specific for Cryptosporidium-derived nucleic acid was carried out as follows. Cryptosporidium **hybridization assay probe** with the base sequence of (A) was synthesized to include a non-nucleotide linker positioned between nucleotides 11 and 12. This **hybridization assay probe** was of the same sense as the Cryptosporidium **target** rRNA and was used to detect the product of a transcription-mediated amplification described by Kacian et al., in U.S. Patent Nos. 5,399,491 and 5,480,784. Ribosomal RNA from C.parvum and C.muris was separately amplified using a promoter-primer with a 5' end promoter base sequence of (S1), a 3' end sense template-specific base sequence of (S2), a primer with an antisense template-specific base sequence of (B), a promoter-primer with a 5' end promoter base sequence of (S1), a 3' end sense template-specific base sequence of (S2), and a primer with an antisense template-specific base sequence of (C). The samples were incubated after amplification. Following a 60 minute incubation at 42 degrees Centigrade in the circulating water bath, 100 microl 1X **Hybridization** Reagent containing 100 fmol of the **hybridization assay probe** was added to each tube, the samples were incubated for 30 minutes at 60 degrees Centigrade in the circulating water bath, and the signal from the annealed **hybridization** assay probes was detected. Sample sets with an average relative light units (RLU) value greater than 10-fold the

average

RLU value for the negative control indicated amplification of the **target** rRNA, and sample sets with an average RLU value less than 10-fold the average RLU for the negative control indicated no amplification of the **target** rRNA. CTATCAGCTTTAGACGGTAGGG

(A) AATTTAATACGACTCACTATAGGGAGA

(S1) CTGCCTTCCTTAGATGTGGTAG

(S2) GCCATGCATGTCTAAGTATAAAC

(B) GGATAACCGTGGTAATTCTAGAG

(C) (133 pages)

using a **helper DNA oligonucleotide**
which **hybridizes** with the **target** in a
different region than the **DNA probe**

PATENT ASSIGNEE: M.L.Technol.Ventures
PATENT INFO: EP 318245 31 May 1989
APPLICATION INFO: EP 1988-311036 22 Nov 1988
PRIORITY INFO: US 1987-124975 24 Nov 1987
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 1989-159301 [22]
AN 1989-10053 BIOTECHDS

AB A new process for **enhancing** binding between a nucleotide **probe** and a complementary nucleotide sequence in single-stranded **target** nucleic acid involves adding a **helper oligonucleotide**, which **hybridizes** with the **target** nucleic acid in a different region than the **probe**, to the **target**. The **target** nucleic acid is DNA, mRNA, rRNA, tRNA or other small nucleic acid. The **probe** (10-50 nucleotides) and the **helper oligonucleotide** (10-100 nucleotides) may be DNA with a diphosphate ester, alkyl, arylphosphate, or phosphorothioate backbone. The **probe** may be labeled with ¹²⁵I or an acridium ester. A new duplex nucleic acid comprises a hybrid of a nucleotide **probe** and a **target** nucleic acid to which a **helper oligonucleotide** is **hybridized**. The **helper oligonucleotide** imposes a secondary and tertiary structure on the targeted region, accelerating the rate of **probe** binding. The **helper oligonucleotide** can also raise melting temp. of the hybrid of a relatively short **probe** and its intended **target**, so that assays for organisms which occur in environments populated by closely related organisms can be obtained with improved specificity. (17pp)

L9 ANSWER 4 OF 10 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.

ACCESSION NUMBER: 2000285313 EMBASE

TITLE: Unlabeled **helper oligonucleotides**
increase the in situ accessibility to 16S rRNA of
fluorescently labeled **oligonucleotide** probes.

AUTHOR: Fuchs B.M.; Glockner F.O.; Wulf J.; Amann R.

CORPORATE SOURCE: B.M. Fuchs, Max-Planck-Inst. fur Mar. Mikrobiol.,
Celsiusstr. 1, D-28359 Bremen, Germany.
bfuchs@mpi-bremen.de

SOURCE: Applied and Environmental Microbiology, (2000) 66/8
(3603-3607).

Refs: 17

ISSN: 0099-2240 CODEN: AEMIDF

COUNTRY: United States

DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 004 Microbiology

LANGUAGE: English

SUMMARY LANGUAGE: English

AB **Target** site inaccessibility represents a significant problem for fluorescence in situ **hybridization** (FISH) of 16S rRNA with **oligonucleotide** probes. Here, unlabeled **oligonucleotides** (helpers) that bind adjacent to the **probe target** site were evaluated for their potential to increase weak **probe hybridization** signals in Escherichia coli DSM 30083(T). The use of helpers **enhanced** the fluorescence signal of all six probes examined at least fourfold. In one case, the signal of **probe** Eco474 was increased 25-fold with the use of a single **helper probe**, H440-2. In another case, four unlabeled helpers raised the FISH signal of a formerly weak **probe**, Eco585, to the level of

the brightest monolabeled **oligonucleotide** probes available for *E. coli*. The temperature of dissociation and the mismatch discrimination of probes were not significantly influenced by the addition of helpers. Therefore, using helpers should not cause labeling of additional

nontarget

organisms at a defined stringency of **hybridization**. However, the **helper** action is based on sequence-specific binding, and there is thus a potential for narrowing the **target** group which must be considered when designing helpers. We conclude that helpers can open inaccessible rRNA regions for FISH with **oligonucleotide** probes and will thereby further improve the applicability of this technique for in situ identification of microorganisms.

L9 ANSWER 5 OF 10 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.

ACCESSION NUMBER: 2000:424485 BIOSIS

DOCUMENT NUMBER: PREV200000424485

TITLE: Unlabeled **helper oligonucleotides**
increase the in situ accessibility to 16S rRNA of
fluorescently labeled **oligonucleotide** probes.

AUTHOR(S): Fuchs, Bernhard M. (1); Gloeckner, Frank Oliver; Wulf,
Joerg; Amann, Rudolf

CORPORATE SOURCE: (1) Max-Planck-Institut fuer Marine Mikrobiologie,
Celsiusstr. 1, D-28359, Bremen Germany

SOURCE: Applied and Environmental Microbiology, (August, 2000)
Vol.

66, No. 8, pp. 3603-3607. print.

ISSN: 0099-2240.

DOCUMENT TYPE: Article

LANGUAGE: English

SUMMARY LANGUAGE: English

AB **Target** site inaccessibility represents a significant problem for fluorescence in situ **hybridization** (FISH) of 16S rRNA with **oligonucleotide** probes. Here, unlabeled **oligonucleotides** (helpers) that bind adjacent to the **probe target** site were evaluated for their potential to increase weak **probe hybridization** signals in *Escherichia coli* DSM 30083T. The use of helpers **enhanced** the fluorescence signal of all six probes examined at least fourfold. In one case, the signal of **probe** Eco474 was increased 25-fold with the use of a single **helper probe**, H440-2. In another case, four unlabeled helpers raised the FISH signal of a formerly weak **probe**, Eco585, to the level of the brightest monolabeled **oligonucleotide** probes available for *E. coli*. The temperature of dissociation and the mismatch discrimination of probes were not significantly influenced by the addition of helpers. Therefore, using helpers should not cause labeling of additional

nontarget

organisms at a defined stringency of **hybridization**. However, the **helper** action is based on sequence-specific binding, and there is thus a potential for narrowing the **target** group which must be considered when designing helpers. We conclude that helpers can open inaccessible rRNA regions for FISH with **oligonucleotide** probes and will thereby further improve the applicability of this technique for in situ identification of microorganisms.

L9 ANSWER 6 OF 10 SCISEARCH COPYRIGHT 2003 ISI (R)

ACCESSION NUMBER: 2000:604507 SCISEARCH

THE GENUINE ARTICLE: 340QP

TITLE: Unlabeled **helper oligonucleotides**
increase the in situ accessibility to 16S rRNA of
fluorescently labeled **oligonucleotide** probes

AUTHOR: Fuchs B M (Reprint); Glockner F O; Wulf J; Amann R

CORPORATE SOURCE: MAX PLANCK INST MARINE MIKROBIOL, CELSIUSSTR 1, D-28359
BREMEN, GERMANY (Reprint)
COUNTRY OF AUTHOR: GERMANY
SOURCE: APPLIED AND ENVIRONMENTAL MICROBIOLOGY, (AUG 2000) Vol.
66, No. 8, pp. 3603-3607.
Publisher: AMER SOC MICROBIOLOGY, 1752 N ST NW,
WASHINGTON, DC 20036-2904.
ISSN: 0099-2240.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE; AGRI
LANGUAGE: English
REFERENCE COUNT: 17

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB **Target** site inaccessibility represents a significant problem for fluorescence in situ **hybridization** (FISH) of 16S rRNA with **oligonucleotide** probes. Here, unlabeled **oligonucleotides** (helpers) that bind adjacent to the **probe target** site were evaluated for their potential to increase weak **probe hybridization** signals in Escherichia coli DSM 30083(T). The use of helpers **enhanced** the fluorescence signal of all six probes examined at least fourfold. In one case, the signal of **probe** Eco474 was increased 25-fold with the use of a single **helper probe**, H440-2. In another case, four unlabeled helpers raised the FISH signal of a formerly weak **probe**, Eco585, to the level of the brightest monolabeled **oligonucleotide** probes available for E. coli. The temperature of dissociation and the mismatch discrimination of probes were not significantly influenced by the addition of helpers. Therefore, using helpers should not cause labeling of additional nontarget organisms at a defined stringency of **hybridization**. However, the **helper** action is based on sequence-specific binding, and there is thus a potential for narrowing the **target** group which must be considered when designing helpers. We conclude that helpers can open inaccessible rRNA regions for FISH with **oligonucleotide** probes and will thereby further improve the applicability of this technique for in situ identification of microorganisms.

L9 ANSWER 7 OF 10 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 2002:812019 CAPLUS

DOCUMENT NUMBER: 137:333989

TITLE: LNA **helper** probes for detection of a single nucleotide polymorphism by a capture **oligonucleotide**

INVENTOR(S): Jacobsen, Nana; Jakobsen, Mogens Havsteen; Skouv, Jan

PATENT ASSIGNEE(S): Exiqon A/S, Den.

SOURCE: Eur. Pat. Appl., 24 pp.

CODEN: EPXXDW

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
EP 1251183	A2	20021023	EP 2002-388014	20020218
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				

PRIORITY APPLN. INFO.: US 2001-284729P P 20010418

AB The invention relates to a method for **enhancing hybridization** of a capture **oligonucleotide** to a **target** sequence using a **helper probe**

comprising modified nucleotide residues. The said capture oligonucleotide or amplicon is conjugated to a reporter group. The method exhibits significantly improved binding abilities of capture oligonucleotide to target sequence. In particular the method is suitable for detection of single nucleotide polymorphism or genotyping a human or animal.

L9 ANSWER 8 OF 10 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 2002:51666 CAPLUS

DOCUMENT NUMBER: 136:97271

TITLE: Dipstick assays for detection and capture of target nucleic acids in solution using helper probes

INVENTOR(S): Lee, Helen; Dineva, Magda Anastassova; Hazelwood, Shaun Christopher

PATENT ASSIGNEE(S): UK

SOURCE: PCT Int. Appl., 69 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2002004668	A2	20020117	WO 2001-GB3024	20010706
<p>W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM</p> <p>RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG</p>				
AU 2001067752	A5	20020121	AU 2001-67752	20010706
<p>PRIORITY APPLN. INFO.: GB 2000-16814 A 20000707</p> <p>WO 2001-GB3024 W 20010706</p>				
<p>AB Use of helper probes in dipstick assays is described. A dipstick assay is used to test for the presence of a target nucleic acid in a sample soln. The sample soln. is contacted with the dipstick contact end to cause the sample soln. to move by capillary action to a capture zone of the dipstick where the target nucleic acid is captured by a capture probe.. A labeled detection probe is provided that hybridizes to one region of the target nucleic acid and contains a detection ligand, while the capture probe which hybridizes to another region of the target nucleic acid contains a capture ligand. Helper probes may be used to enhance the binding of the capture and/or detection probe to the target nucleic acid, thereby improving the sensitivity of target nucleic acid detection. The first helper probe binds to a second sequence of the target nucleic acid which is either adjacent to the first sequence or is spaced up to 10 nucleotides from the first sequence. The second helper probe binds to a third sequence of the target nucleic acid which is either 10 nucleotides away from or adjacent to the first sequence. Hence, the second and third sequences flank the first sequence of the target nucleic acid. The detection probe may comprise a hook detection probe which hybridizes to a fourth sequence of the target nucleic acid and a universal detection probe which</p>				

hybridizes to the hook detection **probe**. Also, a third helper probe which binds to a fifth sequence of a target nucleic acid may be included to **enhance** binding of the detection probe to the fourth sequence of the target nucleic acid. The capture probe may comprise a universal capture probe **hybridized** to a hook capture probe. A complex may be formed between the detection probe, the first helper probe and the target nucleic acid in soln. which moves up to the capture zone by capillary action where the capture probe contacts the target nucleic acid. Alternatively, a quaternary complex may be formed between the target nucleic acid, detection probe, capture probe and helper probe in soln. prior to binding a capture moiety at the capture zone of the dipstick. In other methods, the helper probes, detection probes or capture probe may also be immobilized to the region between the contact and capture zones and released upon binding the target nucleic acid or they may be releasably immobilized at the capture zone. Dipsticks and kits are also described for diagnosis of sexually transmitted diseases like Chlamydia.

L9 ANSWER 9 OF 10 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 2000:552840 CAPLUS

DOCUMENT NUMBER: 134:37611

TITLE: Unlabeled **helper oligonucleotides** increase the in situ accessibility to 16S rRNA of fluorescently labeled **oligonucleotide** probes

AUTHOR(S): Fuchs, Bernhard M.; Glockner, Frank Oliver; Wulf, Jorg; Amann, Rudolf

CORPORATE SOURCE: Max-Planck-Institut fur Marine Mikrobiologie, Bremen, D-28359, Germany

SOURCE: Applied and Environmental Microbiology (2000), 66(8), 3603-3607

CODEN: AEMIDF; ISSN: 0099-2240

PUBLISHER: American Society for Microbiology

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Target site inaccessibility represents a significant problem for fluorescence in situ **hybridization** (FISH) of 16S rRNA with oligonucleotide probes. Here, unlabeled oligonucleotides (helpers) that bind adjacent to the **probe target** site were evaluated for their potential to increase weak **probe hybridization** signals in Escherichia coli DSM 30083T. The use of helpers **enhanced** the fluorescence signal of all six probes examd. at least fourfold. In one case, the signal of probe Eco474 was increased 25-fold with the use of a single helper probe, H440-2. In another case, four unlabeled helpers raised the FISH signal of a formerly weak probe, Eco585, to the level of the brightest monolabeled oligonucleotide probes available for E. coli. The temp. of dissocn. and the mismatch discrimination of probes were not significantly influenced by the addn. of helpers. Therefore, using helpers should not cause labeling of addnl. nontarget organisms at a defined stringency of **hybridization**. However, the helper action is based on sequence-specific binding, and there is thus a potential for narrowing the target group which must be considered when designing helpers. We conclude that helpers can open inaccessible rRNA regions for FISH with oligonucleotide probes and will thereby further improve the applicability of this technique for in situ identification of microorganisms.

REFERENCE COUNT: 17 THERE ARE 17 CITED REFERENCES AVAILABLE FOR THIS

RECORD. ALL CITATIONS AVAILABLE IN THE RE

FORMAT

L9 ANSWER 10 OF 10 CAPLUS COPYRIGHT 2003 ACS
 ACCESSION NUMBER: 1990:154800 CAPLUS
 DOCUMENT NUMBER: 112:154800
 TITLE: **Helper oligonucleotides for enhancing nucleic acid hybridization**
 assays, their use, and a kit containing them
 INVENTOR(S): Hogan, James John; Milliman, Curt Lawrence
 PATENT ASSIGNEE(S): ML Technology Ventures, L. P., USA
 SOURCE: Eur. Pat. Appl., 17 pp.
 CODEN: EPXXDW
 DOCUMENT TYPE: Patent
 LANGUAGE: English
 FAMILY ACC. NUM. COUNT: 1
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
EP 318245	A2	19890531	EP 1988-311036	19881122
EP 318245	A3	19900912		
EP 318245	B1	19940608		
R: AT, BE, CH, DE, ES, FR, GB, GR, IT, LI, LU, NL, SE				
US 5030557	A	19910709	US 1987-124975	19871124
AT 106947	E	19940615	AT 1988-311036	19881122
ES 2056115	T3	19941001	ES 1988-311036	19881122
WO 8904876	A1	19890601	WO 1988-US4103	19881123
W: AU, DK, FI, JP, KR, NO				
AU 8826112	A1	19890614	AU 1988-26112	19881123
AU 613989	B2	19910815		
JP 02502250	T2	19900726	JP 1989-500216	19881123
CA 1319336	A1	19930622	CA 1988-583849	19881123
FI 8903526	A	19890721	FI 1989-3526	19890721
NO 8902990	A	19890913	NO 1989-2990	19890721
DK 8903612	A	19890920	DK 1989-3612	19890721
PRIORITY APPLN. INFO.:			US 1987-124975	19871124
			EP 1988-311036	19881122
			WO 1988-US4103	19881123

GI

5'-CCTCCCCGCTGAAAGTACTTTAC-3' I

5'-GGTGCTTCTTCTGCGGGTAACGTCAATGAG-3' II

AB A process is provided for **enhancing** the binding between a nucleotide **probe** and a complementary nucleotide sequence in a single-stranded **target** nucleic acid; the process comprises adding a **helper oligonucleotide** which **hybridizes** with the **target** nucleic acid in a different region than the **probe**, the **helper oligonucleotide** being added in an amt. effective to **enhance** binding of **probe** and **target** nucleic acid. The **helper oligonucleotide** imposes a different secondary and tertiary structure on the target; the resulting hybrid of probe and **helper**-target hybrid also exhibits a higher melting temp. (T_m) than the hybrid which results from the addn. of the probe alone. Thus, assays for Salmonella enteritidis rRNA were performed using